



UM01531.ST25.txt
SEQUENCE LISTING

<110> Hale, Calvin C
Price, Elmer M

<120> LARGE SCALE EXPRESSION AND PURIFICATION OF RECOMBINANT PROTEINS

<130> UMO 1531.1

<140> US 09/901,419

<141> 2001-07-09

<160> 5

<170> PatentIn version 3.1

C3

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<211> 4087

<212> DNA

<213> Bos taurus

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<222> (268)..(363)

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<221> misc_feature

<222> (3178)..()

<223> A Poly (H) affinity tag comprising 6 His residues have been inserted at the C-Terminus end of the coding region of the protein

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cagagtctcg agatgcgacg ctacaatctg cagttttcac tagcttccca gtaggttggg      240
acagttggaa ctctgccatt gcccagc  atg ctg cag ttc agt ctg tca ccc acc      294
                               Met Leu Gln Phe Ser Leu Ser Pro Thr
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10                               15                               20                               25

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His Val Asp His Ile Ser Ala Glu Thr Glu Met Glu Gly Glu Gly Asn
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Ser Ile Ile Ala Asp Arg Phe Met Ser Ser Ile Glu Val Ile Thr Ser
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Gln Glu Lys Glu Ile Thr Ile Lys Lys Pro Asn Gly Glu Thr Thr Lys
                               110                               115                               120

aca act gtg agg atc tgg aat gag aca gtg tcc aac ctg acc ttg atg      678
Thr Thr Val Arg Ile Trp Asn Glu Thr Val Ser Asn Leu Thr Leu Met
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gcc ctg ggg tct tca gct cca gag att ctc ctt tca gta atc gag gtg      726
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gac Asp 250	agg Arg	agg Arg	ctt Leu	ctg Leu	ttt Phe 255	tac Tyr	aag Lys	tat Tyr	gtc Val	tac Tyr 260	aag Lys	agg Arg	tat Tyr	cgg Arg	gct Ala 265	1062
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aac Asn	acg Thr 395	gaa Glu	gtg Val	gct Ala	gaa Glu	aat Asn 400	gac Asp	cct Pro	gtc Val	agt Ser	aag Lys 405	atc Ile	ttc Phe	ttt Phe	gaa Glu	1494
caa Gln 410	ggg Gly	aca Thr	tat Tyr	cag Gln	tgt Cys 415	ctg Leu	gag Glu	aac Asn	tgt Cys	ggc Gly 420	aca Thr	gta Val	gcc Ala	ctg Leu	acc Thr 425	1542

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ccc tat aag acc att gag ggg acc gcc aga ggt gga ggg gag gac ttt Pro Tyr Lys Thr Ile Glu Gly Thr Ala Arg Gly Gly Gly Glu Asp Phe 570 575 580 585	2022
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C3
CMT

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Phe																	
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<213> Bos taurus

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<223> A Poly (H) affinity tag comprising 6 His residues have been inser

ted at the C-Terminus end of the coding region of the protein

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Ile Ala Met Val Ala Leu Leu Phe Ser His Val Asp His Ile Ser Ala
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35 40 45

Ser Tyr Tyr Cys Lys Lys Gly Val Ile Leu Pro Ile Trp Glu Pro Gln
50 55 60

Asp Pro Ser Phe Gly Asp Lys Ile Ala Arg Ala Thr Val Tyr Phe Val
65 70 75 80

Ala Met Val Tyr Met Phe Leu Gly Val Ser Ile Ile Ala Asp Arg Phe
85 90 95

Met Ser Ser Ile Glu Val Ile Thr Ser Gln Glu Lys Glu Ile Thr Ile
100 105 110

Lys Lys Pro Asn Gly Glu Thr Thr Lys Thr Thr Val Arg Ile Trp Asn
115 120 125

Glu Thr Val Ser Asn Leu Thr Leu Met Ala Leu Gly Ser Ser Ala Pro
130 135 140

Glu Ile Leu Leu Ser Val Ile Glu Val Cys Gly His Asn Phe Thr Ala
145 150 155 160

Gly Asp Leu Gly Pro Ser Thr Ile Val Gly Ser Ala Ala Phe Asn Met
165 170 175

Phe Ile Ile Ile Ala Leu Cys Val Tyr Val Val Pro Asp Gly Glu Thr
180 185 190

Arg Lys Ile Lys His Leu Arg Val Phe Phe Val Thr Ala Ala Trp Ser
195 200 205

Ile Phe Ala Tyr Thr Trp Leu Tyr Ile Ile Leu Ser Val Ser Ser Pro
210 215 220

Gly Val Val Glu Val Trp Glu Gly Leu Leu Thr Phe Phe Phe Phe Pro
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Asp Gly Lys Val Val Asn Ser His Val Asp Ser Phe Leu Asp Gly Ala
290 295 300

Leu Val Leu Glu Val Asp Glu Arg Asp Gln Asp Asp Glu Glu Ala Arg
305 310 315 320

Arg Glu Met Ala Arg Ile Leu Lys Glu Leu Lys Gln Lys His Pro Glu
325 330 335

Lys Glu Ile Glu Gln Leu Ile Glu Leu Ala Asn Tyr Gln Val Leu Ser
340 345 350

Gln Gln Gln Lys Ser Arg Ala Phe Tyr Arg Ile Gln Ala Thr Arg Leu
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Met Thr Gly Ala Gly Asn Ile Leu Lys Arg His Ala Ala Asp Gln Ala
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Arg Lys Ala Val Ser Met His Glu Val Asn Thr Glu Val Ala Glu Asn
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Asp Pro Val Ser Lys Ile Phe Phe Glu Gln Gly Thr Tyr Gln Cys Leu
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Glu Asn Cys Gly Thr Val Ala Leu Thr Ile Ile Arg Arg Gly Gly Asp
420 425 430

Leu Thr Asn Thr Val Phe Val Asp Phe Arg Thr Glu Asp Gly Thr Ala
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Asn Ala Gly Ser Asp Tyr Glu Phe Thr Glu Gly Thr Val Val Phe Lys
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Pro Gly Glu Thr Gln Lys Glu Ile Arg Val Gly Ile Ile Asp Asp Asp
465 470 475 480

Ile Phe Glu Glu Asp Glu Asn Phe Leu Val His Leu Ser Asn Val Lys
485 490 495

Val Ser Leu Glu Ala Ser Glu Asp Gly Ile Leu Glu Ala Ser His Val
500 505 510

Ser Thr Leu Ala Cys Leu Gly Ser Pro Ser Thr Ala Thr Val Thr Ile
515 520 525

Phe Asp Asp Asp His Ala Gly Ile Phe Thr Phe Glu Glu Pro Val Thr
530 535 540

His Val Ser Glu Ser Ile Gly Ile Met Glu Val Lys Val Leu Arg Thr
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580 585 590

Glu Phe Gln Asn Asp Glu Ile Val Lys Thr Ile Ser Val Lys Val Ile
595 600 605

Asp Asp Glu Glu Tyr Glu Lys Asn Lys Thr Phe Phe Leu Glu Ile Gly
610 615 620

Glu Pro Arg Leu Val Glu Met Ser Glu Lys Lys Ala Leu Leu Leu Asn
625 630 635 640

Glu Leu Gly Gly Phe Thr Ile Thr Gly Lys Tyr Leu Tyr Gly Gln Pro
645 650 655

Val Phe Arg Lys Val His Ala Arg Glu His Pro Leu Pro Ser Thr Ile
660 665 670

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675 680 685

Glu Glu Glu Glu Arg Arg Ile Ala Glu Met Gly Arg Pro Ile Leu Gly
690 695 700

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705 710 715 720

Ser Thr Val Asp Lys Leu Ile Lys Lys Thr Asn Leu Ala Leu Val Val
725 730 735

Gly Thr Asn Ser Trp Arg Glu Gln Phe Ile Glu Ala Ile Thr Val Ser
Page 9

C3
Cont

740

745

750

Ala Gly Glu Asp Asp Asp Asp Asp Glu Cys Gly Glu Glu Lys Leu Pro
 755 760 765

Ser Cys Phe Asp Tyr Val Met His Phe Leu Thr Val Phe Trp Lys Val
 770 775 780

Leu Phe Ala Phe Val Pro Pro Thr Glu Tyr Trp Asn Gly Trp Ala Cys
 785 790 795 800

Phe Ile Val Ser Ile Leu Met Ile Gly Leu Leu Thr Ala Phe Ile Gly
 805 810 815

Asp Leu Ala Ser His Phe Ala Cys Thr Ile Ala Leu Lys Asp Ser Val
 820 825 830

Thr Ala Val Val Phe Val Ala Leu Gly Thr Ser Val Pro Asp Thr Phe
 835 840 845

Ala Ser Lys Val Ala Ala Thr Gln Asp Gln Tyr Ala Asp Ala Ser Ile
 850 855 860

Gly Asn Val Thr Gly Ser Asn Ala Val Asn Val Phe Leu Gly Ile Gly
 865 870 875 880

Val Ala Trp Ser Ile Ala Ala Ile Tyr His Ala Ala Asn Gly Glu Gln
 885 890 895

Phe Lys Val Ser Pro Gly Thr Leu Ala Phe Ser Val Thr Leu Phe Thr
 900 905 910

Ile Phe Ala Phe Ile Asn Val Gly Val Leu Leu Tyr Arg Arg Arg Pro
 915 920 925

Glu Ile Gly Gly Glu Leu Gly Gly Pro Arg Thr Ala Lys Leu Leu Thr
 930 935 940

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35      40      45
Ser Tyr Tyr Cys Lys Lys Gly Val Ile Leu Pro Ile Trp Glu Pro Gln
50      55      60
Asp Pro Ser Phe Gly Asp Lys Ile Ala Arg Ala Thr Val Tyr Phe Val
65      70      75      80
Ala Met Val Tyr Met Phe Leu Gly Val Ser Ile Ile Ala Asp Arg Phe
85      90      95
Met Ser Ser Ile Glu Val Ile Thr Ser Gln Glu Lys Glu Ile Thr Ile
100     105     110
Lys Lys Pro Asn Gly Glu Thr Thr Lys Thr Thr Val Arg Ile Trp Asn
115     120     125
Glu Thr Val Ser Asn Leu Thr Leu Met Ala Leu Gly Ser Ser Ala Pro
130     135     140
Glu Ile Leu Leu Ser Val Ile Glu Val Cys Gly His Asn Phe Thr Ala
145     150     155     160
Gly Asp Leu Gly Pro Ser Thr Ile Val Gly Ser Ala Ala Phe Asn Met
165     170     175
Phe Ile Ile Ile Ala Leu Cys Val Tyr Val Val Pro Asp Gly Glu Thr
180     185     190
Arg Lys Ile Lys His Leu Arg Val Phe Phe Val Thr Ala Ala Trp Ser
195     200     205
Ile Phe Ala Tyr Thr Trp Leu Tyr Ile Ile Leu Ser Val Ser Ser Pro
210     215     220

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 Lys Tyr Val Tyr Lys Arg Tyr Arg Ala Gly Lys Gln Arg Gly Met Ile
 260 265 270
 Ile Glu His Glu Gly Asp Arg Pro Ser Ser Lys Thr Glu Ile Glu Met
 275 280 285
 Asp Gly Lys Val Val Asn Ser His Val Asp Ser Phe Leu Asp Gly Ala
 290 295 300
 Leu Val Leu Glu Val Asp Glu Arg Asp Gln Asp Asp Glu Glu Ala Arg
 305 310 315 320
 Arg Glu Met Ala Arg Ile Leu Lys Glu Leu Lys Gln Lys His Pro Glu
 325 330 335
 Lys Glu Ile Glu Gln Leu Ile Glu Leu Ala Asn Tyr Gln Val Leu Ser
 340 345 350
 Gln Gln Gln Lys Ser Arg Ala Phe Tyr Arg Ile Gln Ala Thr Arg Leu
 355 360 365
 Met Thr Gly Ala Gly Asn Ile Leu Lys Arg His Ala Ala Asp Gln Ala
 370 375 380
 Arg Lys Ala Val Ser Met His Glu Val Asn Thr Glu Val Ala Glu Asn
 385 390 395 400
 Asp Pro Val Ser Lys Ile Phe Phe Glu Gln Gly Thr Tyr Gln Cys Leu
 405 410 415
 Glu Asn Cys Gly Thr Val Ala Leu Thr Ile Ile Arg Arg Gly Gly Asp
 420 425 430
 Leu Thr Asn Thr Val Phe Val Asp Phe Arg Thr Glu Asp Gly Thr Ala
 435 440 445
 Asn Ala Gly Ser Asp Tyr Glu Phe Thr Glu Gly Thr Val Val Phe Lys
 450 455 460
 Pro Gly Glu Thr Gln Lys Glu Ile Arg Val Gly Ile Ile Asp Asp Asp
 465 470 475 480

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Ile Phe Glu Glu Asp Glu Asn Phe Leu Val His Leu Ser Asn Val Lys
485 490 495

Val Ser Leu Glu Ala Ser Glu Asp Gly Ile Leu Glu Ala Ser His Val
500 505 510

Ser Thr Leu Ala Cys Leu Gly Ser Pro Ser Thr Ala Thr Val Thr Ile
515 520 525

Phe Asp Asp Asp His Ala Gly Ile Phe Thr Phe Glu Glu Pro Val Thr
530 535 540

His Val Ser Glu Ser Ile Gly Ile Met Glu Val Lys Val Leu Arg Thr
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595 600 605

Asp Asp Glu Glu Tyr Glu Lys Asn Lys Thr Phe Phe Leu Glu Ile Gly
610 615 620

Glu Pro Arg Leu Val Glu Met Ser Glu Lys Lys Ala Leu Leu Leu Asn
625 630 635 640

Glu Leu Gly Gly Phe Thr Ile Thr Gly Lys Tyr Leu Tyr Gly Gln Pro
645 650 655

Val Phe Arg Lys Val His Ala Arg Glu His Pro Leu Pro Ser Thr Ile
660 665 670

Ile Thr Ile Ala Asp Glu Tyr Asp Asp Lys Gln Pro Leu Thr Ser Lys
675 680 685

Glu Glu Glu Glu Arg Arg Ile Ala Glu Met Gly Arg Pro Ile Leu Gly
690 695 700

Glu His Thr Arg Leu Glu Val Ile Ile Glu Glu Ser Tyr Glu Phe Lys
705 710 715 720

Ser Thr Val Asp Lys Leu Ile Lys Lys Thr Asn Leu Ala Leu Val Val

725

735

Gly Thr Asn Ser Trp Arg Glu Gln Phe Ile Glu Ala Ile Thr Val Ser
740 745 750

Ala Gly Glu Asp Asp Asp Asp Asp Glu Cys Gly Glu Glu Lys Leu Pro
755 760 765

Ser Cys Phe Asp Tyr Val Met His Phe Leu Thr Val Phe Trp Lys Val
770 775 780

Leu Phe Ala Phe Val Pro Pro Thr Glu Tyr Trp Asn Gly Trp Ala Cys
785 790 795 800

Phe Ile Val Ser Ile Leu Met Ile Gly Leu Leu Thr Ala Phe Ile Gly
805 810 815

Asp Leu Ala Ser His Phe Ala Cys Thr Ile Ala Leu Lys Asp Ser Val
820 825 830

Thr Ala Val Val Phe Val Ala Leu Gly Thr Ser Val Pro Asp Thr Phe
835 840 845

Ala Ser Lys Val Ala Ala Thr Gln Asp Gln Tyr Ala Asp Ala Ser Ile
850 855 860

Gly Asn Val Thr Gly Ser Asn Ala Val Asn Val Phe Leu Gly Ile Gly
865 870 875 880

Val Ala Trp Ser Ile Ala Ala Ile Tyr His Ala Ala Asn Gly Glu Gln
885 890 895

Phe Lys Val Ser Pro Gly Thr Leu Ala Phe Ser Val Thr Leu Phe Thr
900 905 910

Ile Phe Ala Phe Ile Asn Val Gly Val Leu Leu Tyr Arg Arg Arg Pro
915 920 925

Glu Ile Gly Gly Glu Leu Gly Gly Pro Arg Thr Ala Lys Leu Leu Thr
930 935 940

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